

groups: normoxic control group (control group), normoxia+sRAGE group, hypoxia/reoxygenation (H/R) group (model group), hypoxia/reoxygenation+sRAGE (H/R+sRAGE) group (experimental group). The viability of myocardial cell was detected by MTT. The leakage of lactate dehydrogenase in culture medium (LDH) was detected by colorimetric method. The activity of superoxide dismutase (SOD) was detected by xanthine oxidase. The content of malondialdehyde (MDA) was detected by thiobarbituric acid color method. The intensity of fluorescence was detected by DCFH-DA fluorescent probe combined flow cytometry-Reactive oxygen species (ROS) levels in response; nitrate reductase determination of nitric oxide (NO) levels.

Results: Compared with H/R group, H/R+sRAGE group can improve the myocardial viability (0.0472 ± 0.0021 vs. 0.0199 ± 0.0012), reduce the amount of LDH leakage (0.0174 ± 0.0054 vs. 0.0642 ± 0.0189), increase SOD activity (14.066 ± 1.3819 vs. 10.418 ± 1.3931), lower MDA (1.1312 ± 0.1975 vs. 1.8200 ± 0.1372) and ROS levels (0.9223 ± 0.1259 vs. 1.3368 ± 0.0691) ($P < 0.05$).

Conclusions: sRAGE may act directly on myocardial cells and antagonize the hypoxia/reoxygenation injury, the protective role is related to inhibition of oxidative stress.

GW25-e3405

The Effect of Huoxue Qianyang Recipe on the Myocardial Gene Expression in Insulin Signaling Pathway of "Blood stasis-Yang kang-Phlegma" Hypertensive Rats

lu bo, Li Jianhua, Gui Mingtai, Zhou Xunjie, Tan Yuanyuan, Yao Lei, Fu Deyu
Yueyang Hospital of Integrated Traditional Chinese Medicine and Western Medicine, Shanghai University of Traditional Chinese Medicine

Objectives: To investigate the Huoxue qianyang recipe's influence on GCK, G6PC and Pdk4 genes' expression in the myocardial insulin signaling pathway of "Blood stasis-Yang kang-Phlegma" hypertensive rat models.

Methods: 36 five-week-old spontaneously hypertensive male rats (spontaneously hypertensive rat, SHR) were selected as objects, and were randomly divided into two groups (per ration of 2:1: SHR blanket control group (SHR-C group) and SHR model group) according to the random number table; rats in SHR model group were treated with Aconite Tang via gavage and fat diet for 4 weeks, and then were randomly divided into SHR control group (SHR-M group) and SHR experiment group (SHR-H group); 9 age-matched WKY rats were selected as the normal control group (WKY-C group). The rats' irritability were observed, the blood pressure of tail artery were recorded. 4 weeks later, the fasting glucose, insulin, lipids, blood viscosity, angiotensin II and other indicators were obtained. The HOMA-IR index was calculated. Then the rats were anesthetized, and the hearts were removed for histological sections. The functional genes in myocardial tissue RNA were screened via insulin functional gene chip, and major key genes (GCK gene, G6PC gene and Pdk4 gene) were selected as the target. The level of GCK mRNA, G6PC mRNA and Pdk4 mRNA were quantitatively identified via PCR method, and the protein expression of GCK, G6PC and Pdk4 gene were detected with Western Blot method.

Results: Before the treatment, the three SHR groups' HOMA-IR index were all higher than the WKY group ($P < 0.01$), and there was no difference among the SHR groups ($P > 0.05$). After treatment, SHR-C, SHR-M groups' HOMA-IR index increased than before ($P < 0.01$), and SHR-H group's HOMA-IR index didn't change significantly ($P > 0.05$). Among three SHR groups, SHR-C had the highest HOMA-IR index ($P < 0.01$), and SHR-H had the lowest ($P < 0.01$). WKY-C group had the highest GCK, G6PC mRNA expression ($P < 0.01$), however SHR-M group had the lowest ones ($P < 0.01$). Both mRNA levels showed notable decrease after modeling and notable increase after treatment. SHR-M group had the highest Pdk4 mRNA expression ($P < 0.01$), however other groups had no significant difference ($P > 0.05$). The Pdk4 mRNA increased after modeling and decreased after treatment. GCK protein level in SHR group rats were significantly lower than WKY-C group ($P < 0.05$), SHR-M group had a further decrease than the other two SHR groups. G6PC protein level in SHR group rats were significantly lower than WKY-C group ($P < 0.01$), and SHR-M group had a lower G6PC protein level than SHR-C group ($P < 0.05$). SHR-M group and SHR-H group had significantly higher Pdk4 protein level than SHR-C group, WKY-C group ($P < 0.01$), but there was no significant difference between the two groups.

Conclusions: Huoxue qianyang recipe (huoxue qianyang qutan) may improve insulin resistance through increasing GCK, G6PC gene expression and decreasing Pdk4 gene expression that involved in insulin PI3K signaling pathway.

GW25-e3453

Effect of Salidroside on the myocardial mitochondria function of rats after acute exhaustive

Zhang Longfei, Cui Yujuan, Cao Xuebin
PLA NO.252 hospital

Objectives: Mitochondria are the sites of aerobic respiration, and generally are the major energy production center in eukaryotes. Our work is to research the effect of salidroside (SAL) on the myocardial mitochondria function of rats after acute exhaustive.

Methods: (1) A total of 40 health male Sprague-Dawley rats were randomly divided into four groups ($n=10$ in each group), including sedentary control group, exhausted group, low-dose SAL group, high-dose SAL group. Each group was administered with low-dose SAL (100mg/Kg), high-dose SAL (300mg/Kg) or saline intragastrically for 14 days. Rats were killed immediately after single bout of exhausted exercise (Thomas exhausted standardization) besides sedentary control group which were

killed in resting state during the same period. (2) Myocardium samples were taken to observe histomorphologic changes by light microscope and electron microscope afterwards. (3) ELISA study for concentration of cTnI, CK, MB in plasma and myocardial tissue. (4) The state 3 respiratory ability and state 4 respiratory ability of the myocardial mitochondrial complexes I with glutamate and malate as substrates, state 3 respiratory ability of the myocardial mitochondrial complexes II with succinate as substrate, state 3 respiratory ability of the myocardial mitochondrial complexes IV with TMPD and Ascorbate. were measured by high-resolution respirometry in order to compared the Respiratory Control Ratio (RCR).

Results: (1) Serum cTnI assay results: Compared with control group, there are no significant differences in high-does group ($P < 0.05$), but the exhausted group and low-does group are significant higher ($P < 0.05$). (2) Serum CK assay results: Compared with control group, the high-does group, low-does group and exhausted group are all significant higher ($P < 0.05$). (3) Serum MB assay results: There were no significant difference between high-does group and control group ($P > 0.05$), the low-does group and exhausted group are all significant higher ($P < 0.05$). (4) State 4 respiratory ability of the myocardial mitochondrial complexes I: There are no significant difference in each group ($P > 0.05$). (5) State 3 respiratory ability of the myocardial mitochondrial complexes I: Compared with control group, the high-does group, low-does group and exhausted group are all significant decreased ($P < 0.05$). (6) Respiratory control ratio of the myocardial mitochondrial complexes I: Compared with control group, the high-does group, low-does group and exhausted group are all significant decreased ($P < 0.05$). (7) State 3 respiratory ability of the myocardial mitochondrial complexes II: Compared with control group, There are no significant difference between high-does group ($P > 0.05$), but the low-does group and exhausted group are significant decrease ($P < 0.05$). (8) State 3 respiratory ability of the myocardial mitochondrial complexes IV: Compared with control group, the high-does group, low-does group and exhausted group are all significant higher ($P < 0.05$).

Conclusions: (1) Acute exhaustive exercise cause myocardial damage. High doses of Salidroside can prevent myocardial damage. (2) Acute exhaustive exercise would induce the respiratory rate of mitochondrial complex I, II, IV in state 3. (3) Salidroside can protect myocardium mitochondrial by improve the mitochondrial respiratory function. Furthermore, the protective effects of salidroside, high does is better than the low dose.

GW25-e4140

Expression and effect of TESTIN on atherosclerosis in Rabbits

Zhang Yue, Yuan Meng, Li Guangping
The 2nd Hospital of Tianjin Medical University

Objectives: TES gene is a component of focal adhesions and cell-to-cell junctions located at 7q31.2. Our purpose was to investigate the expression of TES gene, and its relationship with the development of atherosclerosis in rabbits.

Methods: 32 New Zealand rabbits were divided into two groups randomly: control group and high-cholesterol group. The level of lipids was measured before and after 3-months' high-cholesterol intervention respectively. Immuno-histochemistry/fluorescence method was used to detect the deposition of TES protein in aorta tissues in the two groups; real-time polymerase chain reaction (PCR) and Western blot was performed to compare the expression of TES protein in aorta tissues between the two groups. The correlations of TES gene to the development of atherosclerosis were also analyzed.

Results: After the atherosclerotic model established, the level of the serum lipids in high-cholesterol group increased significantly compared with control group, there was statistical difference between the two groups ($P < 0.05$). We found TES protein expressed in the endothelium layer of arteries predominantly. Real-time PCR analysis showed that the mRNA level of TES was markedly reduced by 10-fold in high-cholesterol group compared with control group ($P = 0.015 < 0.05$), and Western blot analysis also showed the protein level was lower in high-cholesterol group ($P < 0.05$).

Conclusions: The expression level of TES is significantly down-regulated in atherosclerosis. It suggests that TES may play a novel role in the development of atherosclerosis.

GW25-e4142

A new and simple method for isolation of the rabbit's coronary artery without using colored latex and the dissecting microscope

Zhang Yue, Yuan Meng, Li Guangping
The 2nd Hospital of Tianjin Medical University

Objectives: The rabbit, a common experimental animal, has been widely used in various studies. However, Rabbit itself being a smaller animal it is more difficult to observe and separate the coronary artery. Commonly adopted methods in the coronary artery separation in rabbit are: Internal filling, acid separation method and the application of microsurgical microscope. Among these former two uses a color solution, and probably influence the result of HE staining. While the use of microscope is a time consuming complex process which needs higher operating technology. Our purpose was to establish a new and simple method for isolation of rabbit's coronary artery.

Methods: Healthy male New Zealand rabbit obtained from Tianjin Aoyide Company, 1.5 years old and weighed 2.5kg, was anaesthetized with 30 mg/kg of 3% Pento-barbital. Under anesthesia, a rapid lateral thoracotomy was performed. After plastic catheters inserted into aorta, 4% formaldehyde was injected into the aorta. In this process, the coronary artery turned white gradually. After 20 minutes, the heart was isolated including the aortic arch. Then a coronary artery guide wire was plugged into

the left and right coronary sinus. Along the wire, the rabbit coronary was isolated completely.

Results: Without the aid of a microscope, this separation method isolated rabbit coronary artery accurately.

Conclusions: This method provides a better way for the separation of coronary arteries. And it won't influence the results of further pathological observation.

GW25-e5261

Myocardial infarction accelerates the activation of systemic and local cellular immunity in STZ-induced type 1 diabetic rats

Dong Zhifeng, Penglong Wu, Wei Zhu, Meng Wei

Department of Cardiology, Shanghai Sixth Hospital, Shanghai Jiaotong University School of Medicine, Shanghai 200233, China

Objectives: Clinically, diabetes is very common in patients hospitalized for acute myocardial infarction (AMI). It is a strong and independent co-morbidity of all-cause mortality and readmission for post-myocardial infarction chronic heart failure (CHF). The central role for monocyte subset accumulation in the heart following AMI and the role of the spleen as monocyte reservoir were all recently demonstrated. However, whether the associated cellular immunity mechanism was involved in AMI with diabetes was unknown.

Methods: We performed the comparison in four separate groups: 1) rats with sham surgically induced myocardial infarction (Ctr, n=10); 2) rats with surgically induced myocardial infarction (MI, n=10); 3) STZ-induced type 1 diabetic rats (DB, n=10); 4) STZ-induced type 1 diabetic rats with surgically induced myocardial infarction (DB+MI, n=10). The parameters of cellular immunity in the heart, spleen and blood were evaluated by flow cytometry and immunohistochemistry etc. In addition, cardiac remodeling and function was also evaluated.

Results: Twelve weeks after the operation, compared with DB or MI rats, DB+MI rats exhibited the following: 1) significantly increased cardiac enlargement, fibrosis and deteriorated cardiac function; 2) significantly increased infiltration of CD₄₄ T cells and the expression of IFN- γ , IL-17 and IL-4 in heart. 3) significantly increased proportion of CD₄₄ T cells and producing-IFN- γ , IL-17 and IL-4 CD₄₄ T cells and a decreased Treg/Th17 ratio in spleen; 4) significantly increased the proportion of producing IFN- γ , IL-17 and IL-4 CD₄₄ T cells and Treg in blood. However the circulating immune complexes (CIC) and IgG did not showed the difference between them.

Conclusions: In this study, MI significantly accelerated cardiac infiltration of CD₄₄ T cell and the spleen and serum activation of CD₄₄ T cell especially its inflammation associated subgroup in STZ-induced type 1 diabetic rats. Systemic and local cellular immunity probably involved in the post-MI CHF progression in diabetes.

GW25-e5274

Lycopene protects endoplasmic reticulum stress-induced apoptosis against neonatal mouse cardiomyocytes hypoxia/reoxygenation injury

Xu Jiqian^{1,2}, Chen Bin², Zhou Zhou¹, Xu Lei¹, Zhang Shuang¹, Yu Zhengping³, Hu Houxiang¹

¹Department of Cardiology, North Sichuan Medical College First Affiliated Hospital, Nanchong, Sichuan, China, ²Department of Anesthesiology, North Sichuan Medical College First Affiliated Hospital, Nanchong, Sichuan, China, ³Department of Occupational Health, Third Military Medical University, Chongqing, China

Objectives: Endoplasmic reticulum (ER) stress induced apoptosis has been implicated as a critical cause in the pathogenesis of myocardial ischemia reperfusion (I/R) injury. Our previous studies demonstrated that lycopene exhibits great pharmacological potential in protecting against the I/R-injury, but whether its effect is mediated through attenuation of ER stress-induced apoptosis remains unclear. The aim of this study was to investigate the effect of lycopene on hypoxia/reoxygenation (H/R) induced ER stress in primary cultured neonatal mouse cardiomyocytes.

Methods: Primary cardiomyocytes were isolated from neonatal C57BL/6 mice and divided into four groups: control, lycopene, H/R, lycopene + H/R. The cultured cardiomyocytes underwent 4h of hypoxia followed by 6h of reoxygenation to achieve H/R model. Cardiomyocytes were pretreated with lycopene (5 μ M) prior to H/R treatment in lycopene + H/R. Cell viability was assessed using CCK-8 assay in each group. AnnexinV-FITC/PI assay was used to evaluate cardiomyocytes apoptosis in the different treatment groups. The expression of GRP78, a widely used marker of endoplasmic reticulum stress, was measured via western blot. The expression of ER-related apoptotic maker of CHOP/GADD153 and caspase-12 was measured by real-time PCR.

Results: Our results demonstrate that the cell viability significantly decreased to 66.30 \pm 4.84% of the control levels following H/R, the cell viability markedly improved in lycopene + H/R (P <0.01). The results from flow cytometer with Annexin V and PI double-staining illustrated that after exposure to H/R, apoptotic percentage significantly increased to 26.42 \pm 2.71% (P <0.01), while that of control and lycopene group were 4.96 \pm 1.51% and 4.69 \pm 1.42%, respectively. In contrast, lycopene + H/R markedly prevented the H/R-induced apoptosis (16.38 \pm 2.12%, P <0.01). Compared to control and lycopene, the expression of GRP78 protein increased more than two-fold in H/R treatment (P <0.01), while the expression of GRP78 protein only increased to 1.46-fold in lycopene + H/R (P <0.01). In addition, H/R treatment evoked a significant increase in GADD153/CHOP mRNA compared to control groups (P <0.01). However, the GADD153/CHOP mRNA expression was markedly down-regulated to

1.68-fold of control levels with lycopene pretreatment (P <0.01). Furthermore, the caspase-12 mRNA expression was also significantly increased in H/R treatment (1.82 folds of control group, P <0.05). However, pretreatment with lycopene efficiently reduced caspase-12 mRNA expression caused by H/R treatment.

Conclusions: These findings reveal that lycopene protects against H/R injury by attenuation of ER stress and ER stress induced apoptosis in primary cultured neonatal mouse cardiomyocytes; the protective effect of lycopene on cardiomyocytes highlights the therapeutic potential of plant-derived antioxidants against I/R-injury.

GW25-e5288

Non-antiplatelet effect of Clopidogrel: Improving endothelial function in Chinese healthy subjects with different CYP2C19 genotype

Yinzhuan Zhang, Chen Bilian

Xiangya Hospital, Central South University

Objectives: Clopidogrel is demonstrated to improve endothelial function in vitro and in patients with coronary artery disease (CAD). But it remains unclear whether this effect of clopidogrel is associated with CYP2C19 polymorphisms which determining antiplatelet effect of clopidogrel.

Methods: After genotyping, 12 healthy subjects were enrolled in our study. Among them, 6 subjects were CYP2C19*1/*1 (extensive metabolisers, EMs) and the other 6 subjects were CYP2C19*2/*2or*3 (poor metabolisers, PMs). All subjects received 300mg clopidogrel orally. Endothelial function was assessed by measurement of flow-mediated dilation (FMD) of the brachial artery and ADP-induced platelet aggregation was determined using optical aggregometry before and 4h, 24 h after administration of 300mg clopidogrel.

Results: FMD was significantly higher at 4h and 24h after a loading-dose administration of clopidogrel in both CYP2C19 EMs and PMs groups, which showed no significant difference between the two groups. ADP-induced platelet aggregation was greatly inhibited at 4h and 24h after administration of clopidogrel in CYP2C19 EM group. However, there was no statistical correlation between the change in FMD and ADP-induced platelet aggregation in the two CYP2C19 groups.

Conclusions: It is the first time to report that clopidogrel improves endothelial function in healthy Chinese subjects, which is unrelated with CYP2C19 genotype and independent of antiplatelet action.

GW25-e0839

Incidence of Acute Mountain Sickness in Young Adults at 3200

Meters-Comparison of the Lake Louise Scoring and Chinese Scoring Systems

Chenguo Zhu, Hauglan

Cardiovascular Department of Xinqiao Hospital, Third Military Medical University

Objectives: The purpose of this study was to compare two scoring systems used for diagnosis of acute mountain sickness (AMS): Lake Louise Scoring (AMS-LLS) and Chinese Scoring System (AMS-CSS).

Methods: 339 healthy young adult volunteers, resided at sea level (mean \pm SD: age 24.59 \pm 3.27 years; height 173.93 \pm 5.18 cm; weight 68.21 \pm 7.79 kg), ascended to 3200 m by train and bus, a total journey time of 48 hours, all the persons were ascend as same way, and were divided into three groups. Group 1 (n = 88), group 2 (n = 91) and group 3 (n = 160) were assessed after one, two and three nights, respectively, at altitude.

Results: The overall incidence of AMS was 17.11% (n = 58) and 29.79% (n = 101) according to AMS-LLS and AMS-CSS, respectively. Two participants (0.59%) experienced high altitude pulmonary edema. Both scoring systems showed the highest incidence of AMS after the second night at high altitude. There was a good correlation between AMS-CSS and AMS-LLS scores (Pearson = 0.820, P < 0.001). AMS-CSS identified all AMS subjects diagnosed by AMS-LLS, plus an additional 43 missed by AMS-LLS. The dominant symptoms were reduced exercise tolerance (61.7%), fatigue (49.05%), dizziness (28.9%), chest distress (28.3%) and headache (27.4%). Compared with AMS-LLS, the sensitivity, specificity, and positive and negative predictive values of AMS-CSS were 100%, 84.7%, 57.43% and 100%, respectively. There was no relationship between oxygen saturation (SpO₂) levels and AMS scores at 3200 m.

Conclusions: AMS-CSS is similar, but a little different details, with AMS-LLS. AMS positive diagnosis outnumbers the LLS standard, but there might be a false positive. Headache was not the dominant symptom at 3200 m high altitude in this study, and SpO₂ levels did not correlate with AMS scores.

GW25-e1071

Interleukin-1 beta overexpression in hypothalamic paraventricular nucleus deteriorates heart failure

Liu Qiang, Wang Tao, Yu Huapeng, Liu Bin, Jia Ruyi

Department of Cardiology, The Fourth People's Hospital of Jinan, Medical School, Tai Shan Medical College

Objectives: To investigate whether interaction between interleukin-1 β and angiotensin II receptor 1 in the PVN contributes to progression of HF.

Methods: Rats were implanted with bilateral paraventricular nucleus (PVN) cannulae and subjected to coronary artery ligation or sham surgery (Sham). Subsequently, animals were treated for 4 weeks through PVN infusion with either vehicle, losartan (LOS, 200ug/d), IL-1 β (IL, 1ug/d) or IL-1 β together with losartan (LOS+IL).